

Possible Anticipation in Hereditary Spastic Paraplegia Type 4 (SPG4)

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ABSTRACT: Objective: We report a multigenerational family with uncomplicated hereditary spastic paraplegia type 4 and apparent anticipation. Genetic analysis of the proband revealed a frame shift mutation (5 base pair deletion) in exon 9 of the SPG4 gene encoding the spastin protein. We hypothesized that this deletion mutation may be dynamic and variability in the size of the deletion could account for the anticipation. **Methods:** Clinical and genetic analysis of this family and the deletion mutation. **Results:** In this family, the age of onset, which ranges from 3 to 50 years shows an average decrease in the age of onset of 21.8 years per transmission over three generations. Genetic analysis of multiple family members indicates that all affected members carry the same c.1340_1344delTATAA mutation and that it is not dynamic. **Conclusion:** In this family, other molecular mechanisms may contribute to development of anticipation.

RÉSUMÉ: Possibilité d'anticipation dans la paraplégie spastique héréditaire de type 4 (PSG4). Objectif : Nous rapportons le cas d'une famille de plusieurs générations atteinte de paraplégie spastique héréditaire (PSH) de type 4 non compliquée et ce qui semble être un phénomène d'anticipation. L'analyse génétique chez le cas index a montré une mutation à trame décalée (délétion de 5 paires de bases) dans l'exon 9 du gène SPG4 qui code la synthèse de la spastine. Nous avons émis l'hypothèse que cette délétion puisse être dynamique et que la variabilité de la taille de la délétion puisse être responsable d'un phénomène d'anticipation. **Méthodes :** Analyse clinique et génétique de cette famille et de sa mutation. **Résultats :** Dans cette famille, on note une diminution moyenne de 21,8 ans par transmission sur trois générations, l'âge de début variant de 3 à 50 ans. L'analyse génétique effectuée chez plusieurs membres de la famille indique que tous les individus atteints sont porteurs de la même mutation c.1340_1344delTATAA qui n'est pas dynamique. **Conclusion :** Il est possible qu'un autre mécanisme moléculaire contribue à l'anticipation observée dans cette famille.

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Hereditary spastic paraplegia (HSP) refers to a genetically and clinically heterogeneous group of disorders with a prevalence ranging from 2.0 to 9.6/100,000.^{1,2} One form of HSP transmitted in an autosomal dominant fashion and unassociated with other clinical features has been genetically linked to 2p21-p22.³ This disorder, spastic paraplegia type 4 (SPG) is caused by mutations in the SPG4 gene and is the most common genetic cause of HSP.^{4,5}

Anticipation is a biological phenomenon in which a genetic disorder becomes increasingly severe or presents at an earlier age in successive generations of a family. It is well recognized and has been described in a number of disorders including Huntington's disease, myotonic dystrophy type 1, spinocerebellar ataxia type 1 and dentatorubral pallidolusian atrophy.⁶ All of these conditions involve triplet repeat expansions and the genetic basis of anticipation has been ascribed to the mutational instability of these repeats during intergenerational transmission.

Whether or not anticipation occurs in HSP has been debated.⁷⁻¹⁰ We present a family with a genetically confirmed SPG4 mutation and apparent anticipation.

CASE REPORT

Clinical

Case 1 (III-1, Figure) is a 48-year-old woman who was evaluated for complaints of unsteady gait and weakness in her

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legs which began in the third decade and slowly progressed. She is a nurse and an excellent historian. She recalls that in her teens she was very athletic, a fast runner and had no neurological symptoms. However, following delivery of her son (Case 3) at age 24 years, she began to develop symptoms of spastic paraplegia. Over the ensuing 24 years, her imbalance and stiffness progressively worsened. Family history revealed that multiple members of the family had similar complaints (Figure). The remainder of the neurological review of systems was negative with no complaints of numbness, tingling, dysarthria, diplopia or difficulties with her bowel or bladder. Clinical evaluation showed a normal mental status, cranial nerve and sensory examination. Motor testing was normal except for mild hip flexion weakness. Her deep tendon reflexes were pathologically brisk in both arms and legs with sustained clonus at the ankles. The plantar reflexes were extensor bilaterally and she had a spastic gait.

Case 2 (III-3, Figure), a 51-year-old brother of the index, patient was evaluated for spastic paraparesis which he had for more than 25 years. In his teens and early twenties, like his sister, he was asymptomatic. He had worked as a roofer, an occupation which required agility and balance. He developed difficulties in his mid-twenties and over the ensuing time period, his condition progressed. He stopped working at age 30 years and currently walks with a cane. Neurological examination showed intact mental status and a normal cranial examination. Motor examination revealed normal power and mild weakness of hip flexors with no sensory disturbance. The reflexes were brisk in both extremities with four to five beats of clonus at the ankles and bilateral up going toes. His gait was spastic, narrow based with notable scissoring. He has difficulty with tandem walking and cannot walk with out support.

Case 3 (IV-1, Figure), is 23-years-old and the only child of index patient. His mother noticed that when he first began to walk, his gait was clumsy. Further evaluation was performed at age three and he was diagnosed with spastic paraplegia. As he grew older, he was unable to keep up with any physical activity with his peers and his symptoms worsened. He had no other neurological symptoms. Neurological examination showed an intact mental status and normal cranial nerve examination. Motor testing showed normal power except for mild hip flexion weakness and a normal sensory examination. His deep tendon reflexes were pathologically brisk in both arms and legs with sustained clonus at the ankles. The plantar reflexes were extensor bilaterally and he had a spastic gait.

An independent history was acquired from at least two family members to determine whether other family members were affected and their age of onset. Two members of generation II are affected with the age of onset in their fifties (Figure, II-1, II-2). In addition, another member of generation IV is affected with age of onset in the first decade (Figure, IV-6). Although there is history available that a member of generation I (I-2, Figure) was affected in her 70's, her age of onset is unknown. There is no reliable family history of affected individuals earlier than generation I.

Genetic analysis

Direct testing of highly purified DNA samples was performed by polymerase chain reaction amplification followed by

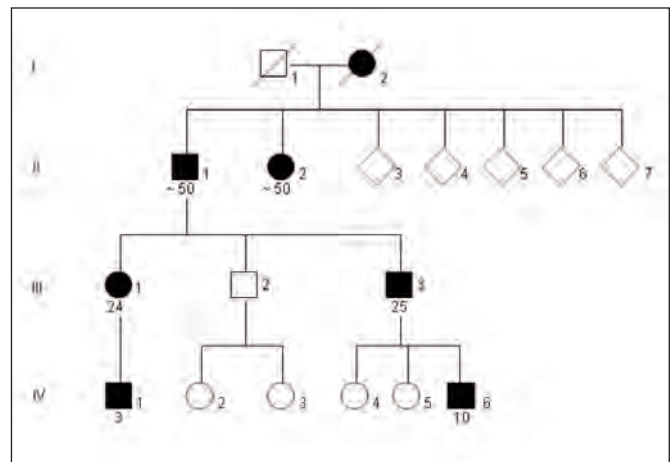


Figure: Pedigree of family with *c.1340_1344delTATAA* mutation with 7 affected members in four generations. The age of onset of spasticity is indicated. Squares represent males, circles females, diamonds unknown and black fill indicate affected individuals.

automated sequencing of both genomic DNA strands for the entire coding region, including the highly conserved flanking intronic sequences of the exon-intron splice junctions (e.g., GT/AG) for all exons in the SPG4 gene. Analysis of the SPG4 gene in this family revealed that cases III-1, III-3 and IV-1 all carry the same 5 base pair deletion (*c.1340_1344delTATAA*) mutation in exon 9 of the SPG4 gene. The DNA sequencing analysis was performed by Athena Diagnostics, Inc. (Worcester, Massachusetts, USA).

DISCUSSION

The affected members of this family have the typical neurological manifestations of uncomplicated spastic paraparesis with an autosomal dominant pattern of inheritance (Figure). Genetic analysis has identified a 5 base pair deletion (*c.1340_1344delTATAA*) mutation in the SPG4 gene. This mutation is expected to shift the reading frame of the SPG4 mRNA resulting in premature protein truncation. Spastic paraplegia type 4 normally possesses 17 exons. This deletion occurring early in the coding sequence of the gene may result in a spastin protein that lacks two of the three highly conserved functional domains, the Walker motif B and the AAA minimal consensus sequence leading to loss of functional protein.¹¹ It is unknown if this mutant spastin protein is stable, or is degraded immediately upon translation. In either case, the spastin protein is unlikely to be functional and therefore this mutation is a loss-of-function mutation leading to the above described phenotype. Previously reported deletion mutations of the SPG4 gene are distributed throughout the gene and account for approximately 23% of all spastin mutations. This is the fourth family that has been reported with this specific mutation.⁵

Genetic anticipation with an earlier age of onset has been observed in some families with familial spastic paraparesis. Prior to the discovery of the SPG4 gene, a number of articles had reported the possible existence of anticipation in SPG4 linked

families.¹²⁻¹⁴ There have also been some reports with the discussion of anticipation in families with specific mutations in this gene.¹⁵⁻¹⁸ Although ascertainment bias must always be a consideration in any discussion of anticipation, we believe that in this family, there is strong historical clinical evidence to support its presence. In this family, the age of onset ranges between 3 to 50 years with an average of earlier age of onset of 21.8 years per generation (Figure). The genetic analysis demonstrates that the c.1340_1344delTATAA mutation is not dynamic and does not change with transmission from one generation to the next. However, there are other molecular mechanisms which could contribute to the development of anticipation. These include the presence of intronic DNA repeat sequences that are often highly mutable and, depending upon repeat size, could influence gene expression to varying degrees. Alternatively, other genes could act as modifiers of the SPG4 gene and ultimately contribute the variability in age of onset.

Further studies of patients and families with genetically confirmed SPG4 mutations are needed to confirm whether or not anticipation exists in this disorder. Confirmation of the presence of anticipation may have implications for genetic counseling and may facilitate fundamental research which could provide insight into the pathophysiology of HSP.

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